

USARIEM TECHNICAL REPORT T-03/7

**Carbohydrate Supplementation Improves Time-Trial Cycle
Performance at 4300 m Altitude**

Prepared
by

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The investigators have adhered to the policies for protection of human subjects as prescribed in Army Regulation 70-25, and the research was conducted in adherence with the provisions of 45 CFR part 46.

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BACKGROUND

The data within this report were collected in the third year (2002) of a three-year collaborative research project between the Palo Alto Veterans Health System (PAVA, Palo Alto, CA) and the U.S. Army Research Institute of Environmental Medicine (USARIEM, Natick, MA). The entire project "*Effect of Energy Deficit on Work Performance at 4,300 m Elevation*" was funded by a grant awarded to the PAVA and the USARIEM by the Cooperative VA/DoD Medical Research Program in the area of physiological foundations of physical performance and combat readiness.

The overall objective of the project was to assess the effects of moderately severe energy imbalance at altitude on physical performance, fuel metabolism, and altitude acclimatization. During the first year of the project, we studied the effects of energy intake deficit (~1,500 Kcals/day) at altitude on physical performance. One of our major findings was that significant losses of lean body mass due to underfeeding did not impair maximal or submaximal physical performance either at sea level or during three weeks of residence at 4,300 m altitude. During year two, additional data from that study and from other related studies within the overall project were also assessed to determine where the scientific effort should best be expended in final year.

It was decided that the reduced energy balance focus be continued in year three. During a two-week residence to 4,300 m, we would produce approximately the same level of energy deficit (i.e., ~1,500 Kcals/day) in all volunteers as in year one. However, the energy deficit would be produced by increasing energy expenditure and not by reducing energy intake. Doing so would allow direct comparisons of a large number of similar measurements between years one and three. Major goals of the final year of the project were to determine the importance of increased daily physical activity on the incidence and severity of Acute Mountain Sickness (AMS) and development of oxidative stress; and to evaluate the effect of antioxidant supplementation (a mixture of β -carotene, α -tocopherol, ascorbic acid, selenium, and zinc) on AMS incidence and severity, oxidative stress, altitude acclimatization and markers of immune function. Another major goal, presented in this report, are the results of a study that determined if carbohydrate supplementation during prolonged exercise would improve exercise performance at altitude.

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EXECUTIVE SUMMARY

Carbohydrate supplementation (CHOS) during prolonged (>1 hr) heavy cycle exercise at sea level (SL) enhances glucose availability and oxidation, allowing performance at higher work rates compared to control. However, at ALT, hypoxemia exacerbated by exercise may limit work rate increases and time-trial cycle performance improvements with CHOS. The purpose of the study was to determine if CHOS improves performance at ALT. After an overnight fast, 2 groups of 8 men, matched by age (~25 yr), maximal oxygen uptake ($\text{VO}_{2\text{max}}$) (~56 ml/kg/min), weight (~78 kg), and height (~179 cm), performed a two-segment endurance test at SL and while living at ALT on days 3 (ALT3) and 10 (ALT10). In segment one, all fasting subjects performed 20 min of steady-state exercise at 48% and 68% their altitude-specific $\text{VO}_{2\text{max}}$. For segment two, they performed a maximum effort, 720 KJ time trial. At the start of the time-trial, and every 15 min thereafter, 8 men (CHO) drank a 10% CHO solution (0.7 g/kg bw) and 8 men (PLA) drank a placebo (double-blind). All freely adjusted work rate (W) and drank water at the start of and throughout the time trial. Blood glucose and O_2 saturation (SaO_2) were measured during the steady-state segment and at rest, 180 KJ, 360 KJ, and 720 KJ during the time-trial. Results ($\text{X} \pm \text{SE}$): During the steady-state segment, blood glucose and SaO_2 responses for both groups were similar for all test days ($P > 0.05$). During the time trial segment at SL, cycle time was similar for both groups (~59 min, $P > 0.05$). At ALT, time was worse than at SL for each group ($P < 0.01$) but was better for CHO than for PLA on ALT3 (80 ± 7 vs 105 ± 9 min, $P < 0.01$) and ALT10 (77 ± 7 vs 90 ± 5 min, $P < 0.01$). Mean work rate during the time trial at SL was similar for both groups (~218 W, $P > 0.05$). At ALT, work rate for both groups was reduced from SL ($P < 0.01$) with the reduction larger for PLA than for CHO on day 3 ($P < 0.04$). Blood glucose at SL and ALT was similar for both groups at rest (~92 mg%, $P > 0.05$) but was 14 to 30 mg% higher for CHO than for PLA during time-trial exercise ($P < 0.01$). On ALT3, SaO_2 fell similarly from rest to exercise for both groups (~86% to ~73%, $P < 0.05$) but on ALT10, SaO_2 fell more for CHO than for PLA (91 ± 1 to $76 \pm 2\%$ vs 90 ± 1 to $81 \pm 1\%$, $P < 0.02$). Conclusion: Superior time-trial cycle performance of CHO vs PLA on ALT3 and ALT10 implies that increased availability of glucose as a metabolic fuel can greatly offset performance impairments linked to hypoxemia exacerbated by exercise during ALT residence in similarly acclimatized men.

INTRODUCTION

Muscle glycogen and blood glucose play critical roles in providing energy during exercise. At the start of prolonged and strenuous exercise at sea level, the required energy is derived approximately equally from fat and carbohydrate, with most of the carbohydrate oxidation coming from muscle glycogen stores (12). As the exercise bout proceeds, the relative contribution to total carbohydrate oxidation by blood glucose increases progressively while that provided by muscle glycogen decreases progressively. Since the primary source of blood glucose (i.e., liver glycogen) is limited and blood glucose utilization is related curvilinearly to exercise intensity (8), endurance performance at high exercise intensities will be impaired if blood glucose level falls (7; 12; 17). Ingesting carbohydrate during prolonged and strenuous exercise maintains blood glucose levels, increases carbohydrate availability, enhances glucose oxidation by active muscle (24), and typically improves endurance performance whether or not blood glucose availability and total carbohydrate oxidation were limiting to performance during control or placebo trials (8; 9; 24).

The effect of carbohydrate supplementation during exercise on endurance performance at altitude has not been studied. Some of the physiological responses and adaptations associated with exercise at altitude make it difficult to determine whether carbohydrate supplementation could improve endurance performance similarly as at sea level. On the one hand, recent studies measuring free fatty acids, glycerol uptake and release, and glucose turnover indicate that during exercise at 4300m altitude there is decreased reliance on fat as a substrate and increased dependence on blood glucose utilization, compared to sea level (5; 25; 26). Since glucose is transported via facilitated diffusion down a concentration gradient (20), and glucose is the most oxygen efficient fuel under conditions of acute and chronic hypoxia (26), it would seem reasonable to suggest that carbohydrate supplementation during prolonged exercise would be at least as beneficial for performance at altitude as at sea level. Carbohydrate supplementation at altitude, like at sea level (8), would be expected to maintain or increase blood glucose levels, carbohydrate availability, and glucose oxidation during prolonged exercise. Such changes would be expected to result in, for example, an increase in exercise intensity (6) such that a fixed amount of work (e.g., "time-trial" performance test) could be completed more quickly than without carbohydrate supplementation (21; 23).

On the other hand, the significant hypoxemia associated with intense exercise at altitude may prevent an increase in exercise intensity resulting from carbohydrate supplementation. At 4300 m altitude, arterial oxygen saturation during rest is reduced compared to sea level (14) and is reduced further during exercise in proportion to increases in exercise intensity (30). Oxygen transport to tissues during low to moderate exercise intensities is maintained at altitude, however, by compensatory increases in cardiac output and/or arterial oxygen capacity (27; 28). Assuming that there exists the *potential* to perform at a higher exercise intensity at altitude by virtue of increased glucose availability (as at sea level), the compensatory changes in cardiac output and/or arterial oxygen capacity may not be adequate to maintain oxygen transport at the higher intensity. A higher cardiac output would also shorten pulmonary capillary mean transit time and exacerbate an already present altitude-induced oxygen diffusion limitation and result in more severe hypoxemia (31). Time at the higher intensity, therefore, may be limited and exercise performance during a prolonged time-trial may not improve significantly. Time at the higher intensity, therefore, may be limited and exercise performance during a prolonged time-trial may not improve significantly.

The purpose of this study was to determine if carbohydrate supplementation during exercise at altitude will improve time-trial cycle performance. It was hypothesized that carbohydrate supplementation will improve time-trial cycle performance compared to placebo despite significant hypoxemia.

METHODS

VOLUNTEERS

Volunteers were recruited during the months of January through April 2002 from advertisements and fliers placed in local newspapers and universities in and around the Palo Alto/San Jose, CA area. Inclusion criteria included being: an active 21 to 35 year old, non-smoking male¹, normal weight for height (body mass index = 20-27), weight stable for 6 months, no metabolic disease, ability to perform strenuous cycle exercise for 1 hour at 75%

¹ Women were restricted from participation because estrogen (which has antioxidant properties) had the potential to confound results of another major sub-study that evaluated the effects of antioxidant supplementation on oxidative stress linked to exercise at high altitude.

of their age-predicted maximal heart rate, perform a single arm curl with a 50-lb weight, and not born or residing within the previous three months at altitudes greater than 2000 m.

Over 700 men initially inquired via phone or e-mail; of these, approximately 60 came to the laboratory located at the Clinical Studies Unit at the Palo Alto VA Hospital for a verbal briefing and facility tour to become familiarized with equipment, procedures, and personnel. Men with evidence of any physical, mental, and/or medical conditions that would make the proposed studies relatively more hazardous were excluded. Additional screening was performed consisting of a physical exam, and routine nutritional, blood, and urine analyses and assessments. If the remaining men were still interested in participating after being fully informed of all aspects of the study, they were asked to provide written consent. Thirty-two men ultimately volunteered to participate and were scheduled for testing at sea level and altitude.

STUDY OVERVIEW

The volunteers were studied at sea level from April through May. All were fed a controlled and well-balanced diet for 7 days ("stabilization phase") to attain energy and nitrogen balance, and body weight stability. During the stabilization phase, a preliminary cycle ergometer maximal oxygen consumption ($\dot{V}O_{2\max}$) test was conducted on day 2 and a preliminary cycle ergometer endurance test was conducted on day 4. On days 8 to 12 at sea level ("baseline phase"), volunteers lived in the Clinical Studies Unit. During this time, they were asked to lose body weight by increasing their energy expenditure by 1500 Kcals each day and to maintain energy intake, relative to the stabilization phase. During the sea-level baseline phase, a definitive cycle $\dot{V}O_{2\max}$ test was conducted on day 2 and a definitive cycle endurance test was conducted on day 3.

In about a six-week period from the beginning of the sea level, baseline-testing phase to the beginning of the altitude-testing phase, 14 of the 32 volunteers voluntarily withdrew from the project mostly due to unanticipated scheduling conflicts with new jobs or school classes. Of these 14, seven had not participated in either sea-level endurance test. The specific deployment schedule for the remaining 18 volunteers subsequently studied was based primarily on their availability to participate at altitude.

Every one to three days between the beginning of July and the beginning of August either one or two volunteers were flown to Colorado Springs, CO (1,800 m) where they spent the first afternoon and night in an apartment while breathing supplemental oxygen supplied

by an oxygen concentrator to maintain sea level oxygen saturation levels (i.e., >96%). At 0530 h the next morning, they were driven in about an hour to the USARIEM High Altitude Research Laboratory at the summit of Pikes Peak (4,300 m) while breathing from masks connected to small oxygen tanks. Immediately after arriving on the summit (day 1), one volunteer removed his mask, and within an hour, began a 1.5 to 4 hour oxidative stress cycle exercise test (procedures and results to be presented in other reports). If there were two volunteers, the second volunteer removed his mask at the start of exercise of the first volunteer, and about an hour later, began his oxidative stress cycle exercise test. On days 2 and 9 of altitude residence, volunteers performed a cycle $\text{VO}_{2\text{max}}$ test and on days 3 and 10, a cycle endurance test. Volunteers lived on the summit a total of 14 days.

While living on the summit, all volunteers were asked to increase their energy expenditure by 1500 kcals/day more than what was required to maintain body weight during the stabilization phase at sea level; energy intake was to be maintained at sea level amounts. To facilitate the increased energy expenditure, a large tent (18'W x 36'L x 8.5'H) was set up at the summit in front of the laboratory where the volunteers had unlimited access to treadmills, cycle ergometers, rowing and ski machines, and lift weights. The volunteers could also play basketball, soccer, and hacky sack, or play catch with a football, baseball, or frisbee. Over the 14 days, volunteers also participated (weather permitting) in 1 to 3 hikes lasting from 1 to 4 hours. The trails were within 5 miles from the summit and were more than 3600 m elevation.

ENERGY INTAKE

Volunteers were fed a standardized diet throughout the study consisting of whole foods and liquid supplements provided in individualized amounts. Energy intake was initially estimated during one of the screening sessions using the Harris-Benedict equation (16), corrected for typical activity level estimated by volunteer recall. Later, during the sea level stabilization phase, energy intake was adjusted to maintain body weight (which was measured daily). Protein content of the diet was held constant (1.2-1.3 g/kg b.w./day) while energy intake was adjusted by adding or subtracting fat and carbohydrate containing foods so that the ratio of these nutrients remained approximately 1:2, respectively. The diet provided approximately 13% protein, 23% fat, and 63% carbohydrate. The diet also contained at least 80% of the Recommended Daily Allowance for vitamins and minerals, but

portions of foods high in antioxidants were strictly regulated throughout the study to satisfy some requirements of the oxidative stress sub-study.

ENERGY EXPENDITURE

Daily energy expenditure was estimated from self-reported activity logs. Each volunteer recorded in 15 min segments all activities for each 24-hour period (midnight to midnight) throughout most of the study. During the baseline phase at sea level and for the first 12 days at altitude, volunteers were asked to increase their daily energy expenditure by 40% (approximately 1500 Kcals/day) above their sea level, body weight maintenance levels determined during the stabilization phase. Their activities were analyzed by the same investigator throughout the entire study using the American College of Sports Medicine Compendium of Physical Activities (1). On several occasions for each volunteer, heart rate was monitored for some activities (e.g., running, hiking) to more accurately assess energy expended. For all volunteers, total daily expenditures were corrected by actual measurement of basal metabolic rate (via open circuit spirometry) determined during the baseline phase at sea level, and on days 2 to 6 and 10 to 12 at altitude. Volunteers were provided daily continuous feedback on energy expended so that adjustments could be made, if needed.

MAXIMAL OXYGEN UPTAKE

An incremental progressive exercise bout to volitional exhaustion on an electromagnetically-braked cycle ergometer (Sensormedics Co., Model 800s, Yorba Linda, CA) was used to assess $\text{VO}_{2\text{max}}$ at sea level on day 3 of the stabilization phase and day 2 of the baseline phase, and at altitude on days 2 and 9. Volunteers pedaled at 70 to 100 rpm for two min at 50 watts (w), 100 w, 150 w, and then in 30 w increments thereafter until O_2 uptake failed to increase or the volunteer stopped the test despite strong encouragement. During the $\text{VO}_{2\text{max}}$ test, oxygen uptake (via metabolic cart, True Max 2400, ParvoMedics, Salt Lake City, UT) and heart rate (via heart rate watch, UNIQ-CIC, Computer Instruments Corp. Hempstead, NY.) were monitored continuously. Results of the $\text{VO}_{2\text{max}}$ tests were used to: 1. set the low and high work rates during the steady-state exercise segment of each endurance test conducted, 2. determine if the increased activity at altitude would improve $\text{VO}_{2\text{max}}$, 3. determine if the anticipated decrease in body weight would worsen $\text{VO}_{2\text{max}}$, 4. estimate the oxygen costs of the self-selected wattages used during the time-trial performance segment of

the endurance test, and 5. determine the percentage of maximal heart rate used during the endurance tests.

BICYCLE ERGOMETER ENDURANCE TEST

All cycle endurance tests consisted of two distinct segments: a steady-state exercise segment and a time-trial performance segment. The steady-state exercise segment was used to assess physiological changes from sea level to altitude, and those due to altitude acclimatization. The time-trial performance segment was used to assess performance changes from sea level to altitude, and those due to carbohydrate supplementation at sea level and during altitude exposure. Volunteers were required to provide maximum effort during all four time-trial performances. During both segments of each of the sea level and altitude endurance tests, water was provided *ad libitum*. Endurance tests were performed using electromagnetically-braked cycle ergometers (Sensormedics Co., Model 800s, Yorba Linda, CA or Warren C. Collins, Inc., Pedalmate, Braintree, MA)² on four separate days: twice at sea level and on days 3 and 10 at altitude.

During the steady-state exercise segment, volunteers warmed up for 5 min at 50 w and then exercised for 20 min at a low intensity ($48 \pm 5\%$) and 20 min at a high intensity ($68 \pm 5\%$) of their altitude-specific $\text{VO}_{2\text{max}}$ followed by a 5 min rest period to allow bathroom use and “stretching out.” Volunteers were then told to begin the time-trial performance segment in which they were required to complete 720 KJ of total work as fast as possible. Volunteers were allowed at any time during cycling to alter pedaling speed and to adjust work rate by any desired increment. This type of performance test was chosen because of its high test-retest reproducibility and low coefficient of variance (18). Volunteers were provided real time feedback (via computer screen graphics) of total work performed and total work remaining. Volunteers were not informed of any of their time-trial performance times until the study was completed.

All four of the endurance tests were conducted in nearly exactly the same manner. The only exceptions were that for the first endurance test (on day 4 of the stabilization phase) there were no blood samples drawn and, in the time-trial performance segment, in addition to

² Calibrated ergometers (via manufacturers specifications) were also “physiologically” calibrated before the study started. That is, oxygen uptakes were measured for an identical range of power outputs on each ergometer using the same individuals. Oxygen uptakes did not differ between ergometers.

water provided *ad libitum*, water was offered at the exact volume, times, and frequency that the carbohydrate supplement or placebo would be for the remaining three endurance tests. The volunteers were also allowed to finish eating a small snack (< 400 Kcals) more than 1 hour before the mid-morning start of exercise.

The second endurance test was conducted during the baseline phase at sea level, and the third and fourth tests were conducted on days 3 and 10 at altitude, respectively. All three were conducted in the mornings after fasting overnight, placement of a forearm venous catheter, and the drawing of a resting fasting blood sample. If there was only one volunteer scheduled, his test started at approximately 8:30 AM. If there were two volunteers scheduled (the maximum per day), they performed the endurance tests side by side in the same room. One volunteer started at approximately 8:30 AM, and the second volunteer started at approximately 9:15 AM. Carbohydrate supplement or placebo was provided only during the time-trial performance segment. Exercise blood samples were obtained during both the steady-state exercise segment (i.e., while fasting) and the time-trial cycle performance segment (i.e., during placebo or carbohydrate supplementation).

Carbohydrate and Placebo Group Assignment

Just prior to the sea-level baseline phase endurance test, a staff member not directly involved with any exercise testing assigned each of the 18 volunteers to either a carbohydrate supplement (n=9) or placebo (n=9) group for the rest of the study. To satisfy some requirements of all sub-studies of the project, individuals were divided into two groups matched ($P>0.10$) on age, body weight, height, percent body fat, end tidal carbon dioxide, and hypoxic ventilatory response. In addition, to aid in interpretation of the steady state and time-trial segments of the endurance tests at sea level and altitude, volunteers were matched in pairs on sea level $\text{VO}_{2\text{max}}$ (ml/min). Neither the volunteer nor the investigators directly participating in any of the sub-studies knew the group assignment until the entire study was completed.

Carbohydrate and Placebo Supplementation

The carbohydrate (CHO) supplement was a tropical punch flavored blend (22) of maltodextrin (mass/volume, 9%), glucose (2%), and fructose (1%) (Ergo Drink, US Army Soldier Systems Command, Natick MA), previously reported to be highly acceptable (22). Each powdered serving was reconstituted with water to a 10% CHO solution. At the start of

the time-trial performance segment and every 15 min thereafter until completion, volunteers consumed either 0.175 g/kg b.w. (e.g., 80 kg b.w. = 14 g CHO per serving) of reconstituted Ergo Drink or an equal volume of indistinguishable placebo. A staff member not directly involved with the endurance tests mixed the carbohydrate and placebo drinks. The volunteer and the investigators participating in the study remained blinded to the supplement assignment until the entire study was completed. The rate of carbohydrate ingested during exercise (56 grams per hour for an 80 kg b.w.) was within guidelines established by the American College of Sports Medicine (2).

Blood Measures

Blood glucose, lactate, and glycerol (Analox GM7 Micro-Stat, Hammersmith, London, UK), and free fatty acids (Elan Diagnostics, ATAC 8000, Smithfield, RI) were determined at rest, and after 15 min of cycling first at 48% and then at 68% of VO_{2max} during the steady-state exercise segment; and after the volunteer completed 25% (i.e., 180 KJ) and 50% (i.e., 360 KJ) of total work, and every 15 minutes thereafter, and at exercise completion (i.e., 720 KJ) during the time-trial performance segment. Volume of blood withdrawn for each blood sample was 6 to 10 ml. The total amount of blood withdrawn for each endurance test ranged from 50 to 110 ml, depending on the duration of the time-trial performance segment. For any volunteer, the total amount of blood withdrawn for all three endurance tests, spread out over at least two months, did not exceed 300 ml.

Other Measures

During all endurance tests, oxygen saturation via noninvasive finger pulse oximetry (Model N-200, Nellcor, Pleasanton, CA) and heart rate via heart rate watch (UNIQ-CIC, Computer Instruments Corp. Hempstead, NY) were monitored continuously, and ratings of perceived exertion (6 to 20 Borg Scale(4)) were obtained every 5 minutes. Standard respiratory data via metabolic cart (True Max 2400, Parvo Medics Salt Lake City, UT) and blood pressure via manual osculation of arm vessels were collected at rest and within the 10th to 15th minute at 48% and at 68% VO_{2max} during the steady-state exercise segment.

STATISTICS

A two-factor (days X group) or three-factor (days X times X group) analysis of variance with repeated measures on one factor (days) or two factors (days and times), respectively,

was utilized for performance, physiological and blood values comparisons. Post hoc analyses (Neuman-Keuls) were performed when appropriate. Independent t-tests were used to compare specific characteristics (e.g., age, height, etc) between groups. Statistical significance was accepted when $P < 0.05$. All values are expressed as means \pm SE unless otherwise indicated.

RESULTS

One volunteer in the CHO supplement group had severe acute mountain sickness for the first three days of altitude residence and was not able to participate in most testing on those days ---- including the endurance test on day 3 --- but was tested on day 10. Another volunteer, in the placebo group, was tested on day 3 but had to leave the summit for personal reasons on the 6th day. Therefore, the data were analyzed and presented using only the 16 volunteers ($n = 8$ in each group) who participated at sea level and on both the 3rd and 10th day at altitude.

The age, body weight, height, and VO_{2max} of the volunteers at the start of the baseline phase are presented in Table 1. Because of deliberate matching of groups on these variables, there were no differences between groups.

TABLE 1. Age, Body Weight and Height, and Maximal Oxygen Uptake for both Experimental Groups.

| Group: | Age (yrs) | Weight (kgs) | Height (cm) | VO_{2max} (ml/min/kg) | VO_{2max} (ml/min) |
|---------|--------------|-----------------|----------------|----------------------------|-------------------------|
| Placebo | 25.1 \pm 2 | 79.8 \pm 4 | 176.8 \pm 2 | 54.0 \pm 2 | 4270 \pm 178 |
| CHO | 25.3 \pm 2 | 75.1 \pm 2 | 180.6 \pm 2 | 58.4 \pm 2 | 4395 \pm 241 |

N=8, Values are means \pm SE, Placebo = Placebo Group; CHO = carbohydrate supplement group

ENERGY INTAKE AND ENERGY EXPENDITURE

There were no differences in mean daily energy expenditure, energy intake, energy deficit or body weight loss between the CHO supplement and placebo groups at sea level or altitude. During the sea level stabilization and baseline phases, daily energy intake averaged 3879 ± 91 Kcals/day for all volunteers. At sea level, in addition to the considerable time demands placed upon the volunteers by the current research project, all remained fully involved with work, school, and/or family. As a consequence, they were able to increase daily energy expenditure from the stabilization phase to the baseline phase by only 216 ± 67

Kcals/day (3898 ± 163 kcal to 4114 ± 139 Kcals/day, $P < 0.01$) and not the ~ 1500 Kcals/day as planned. As a result, body weight remained stable in both phases at sea level for both groups at $\sim 77.5 \pm 2$ kg.

On the summit, more of the volunteer's time could be focused on the requirements of the research study. Unlimited and convenient access to a wide variety of physical activities resulted daily energy expenditures at altitude being consistently higher than at sea level. Energy expenditure averaged 4567 ± 169 Kcals/day (or ~ 700 Kcals/day more than at sea level, $P < 0.01$) in the first 10 days at altitude. Over the same time at altitude, despite strong encouragement to maintain daily energy intake similar to that at sea level, energy intake was voluntarily reduced to an average of 3140 ± 165 Kcals/day (or ~ 750 Kcals/day less than at sea level, $P < 0.01$). The resulting mean daily energy deficit (~ 1450 Kcals/day) at altitude contributed to the body weight decline from 77.0 ± 2 kg on day 3 to 74.7 ± 2 kg on day 10 ($P < 0.01$). A more detailed description of the energy deficit induced by increased energy expenditure and decreased energy intake will be presented in a later publication.

MAXIMAL OXYGEN CONSUMPTION

Table 2 shows VO_{2max} at altitude expressed as ml/min, ml/min/kg, and as a percent of the sea level VO_{2max} value for the two groups. Maximal oxygen consumption did not differ between groups at sea level or at altitude. For both groups, VO_{2max} declined $\sim 26\%$ from sea level on day 2 ($P < 0.01$) and did not change ($P > 0.05$) with continued exposure ($\sim 24\%$ on day 9). These results indicate that the two groups --- matched on VO_{2max} at sea level --- had similar declines in VO_{2max} resulting from altitude exposure. These results also indicate that for both groups increased physical activity at altitude did not improve VO_{2max} and that the loss of body weight did not worsen VO_{2max} .

TABLE 2. Maximal oxygen consumption (VO_{2max}) at sea level and altitude

| | Day 2 | | | | Day 9 | | | |
|------------|-------------------------|------------|----------------------------|------------|-------------------------|------------|----------------------------|------------|
| | VO_{2max} (ml/min) | SL (%) | VO_{2max} (ml/min/kg) | SL (%) | VO_{2max} (ml/min) | SL (%) | VO_{2max} (ml/min/kg) | SL (%) |
| PLA | 3134 ± 79 | 74 ± 1 | 39.9 ± 2 | 74 ± 1 | 3140 ± 91 | 74 ± 1 | 41.2 ± 2 | 77 ± 2 |
| CHO | 3211 ± 193 | 73 ± 2 | 43.0 ± 2 | 74 ± 2 | 3081 ± 158 | 70 ± 2 | 42.5 ± 2 | 77 ± 3 |

Values are means \pm SE. PLA = Placebo Group; CHO = carbohydrate supplement group; SL = sea level

MAXIMAL HEART RATE

Maximal heart rate was similar at sea level and altitude for both groups (Table 3). Maximal heart rate was lower on day 2 at altitude compared to sea level ($P<0.01$). It was also lower at altitude on day 9 compared to day 2 and sea level ($P<0.01$).

TABLE 3: Maximal heart rate at sea level and altitude

| Group: | Sea Level | Day 2, Altitude | Day 9, Altitude |
|--------------|-------------|-----------------|------------------|
| Placebo | 188 ± 4 | $178 \pm 3^*$ | $157 \pm 4^{**}$ |
| Carbohydrate | 187 ± 3 | $176 \pm 4^*$ | $155 \pm 6^{**}$ |

Values are means \pm SE. $^*P<0.01$ from sea level; $^{**}P<0.01$ from sea level and Day 2, altitude

BICYCLE ERGOMETER ENDURANCE TEST

Volunteer Assignments on Testing Days

At sea level, 10 volunteers were tested individually on separate days (5 from the CHO group and 5 from the placebo group). The remaining six were tested in pairs on three other separate days, with each pair consisting of one volunteer from each group.

On days 3 and 10 at altitude, four volunteers were tested individually on separate days (2 from the CHO group and 2 from the placebo group). The remaining 12 were tested in pairs on six other separate days. On two days, the pair consisted of one volunteer from each group; on two days, the pair consisted of two volunteers from the CHO group; and on two days the pair consisted of two volunteers from the placebo group. None of the pairs was identical between sea level and altitude. All these results are consistent with an assignment of testing days that was based primarily on the "randomness" of volunteer availability.

Steady-State Exercise Segment

Work Rate

Table 4 shows the work rates (watts) used at the low (48% VO_{2max}) and high (68% VO_{2max}) work rates at sea level and altitude. For both groups, work rates were reduced from sea level to day 3 of altitude ($P<0.01$); but the identical work rates were used on both days at altitude. For all volunteers, the low exercise work rate at sea level was used as the high exercise work rate at altitude. (Because VO_{2max} decreased at altitude, reducing work rate

allowed comparisons to be made at similar low and high percentages of $\text{VO}_{2\text{max}}$ at altitude as at sea level.) There were no differences between groups at either the low or high work rates at sea level or altitude.

TABLE 4. Steady-state work rates (watts) at sea level and altitude

| | Sea Level | | Altitude, Day 3 | | Altitude, Day 10 | |
|----------------|-----------|----------|-----------------|----------|------------------|----------|
| | Low | High | Low | High | Low | High |
| Placebo | 144 ± 9 | 208 ± 12 | 94 ± 5 | 144 ± 9 | 94 ± 5 | 144 ± 9 |
| CHO | 141 ± 10 | 202 ± 15 | 94 ± 7 | 141 ± 10 | 94 ± 7 | 141 ± 10 |

Values are means ± SE. Placebo = Placebo Group; CHO = carbohydrate-supplemented group.

% $\text{VO}_{2\text{max}}$

Table 5 shows the low (48% $\text{VO}_{2\text{max}}$) and high (68% $\text{VO}_{2\text{max}}$) exercise intensities of altitude-specific $\text{VO}_{2\text{max}}$ used by each group during the steady-state exercise segment of the endurance test. There was no difference between groups for the exercise intensities at sea level or at altitude. Overall, the percentages used on both days at altitude were slightly higher than those at sea level ($P < 0.05$). The difference related to small errors in estimating *a priori* at sea level the exact decline in percentage of $\text{VO}_{2\text{max}}$ that each volunteer would likely experience at altitude. However, at altitude, when each volunteer used the identical work rate for the low and high intensities on both altitude test days, there was no difference between days for either group.

TABLE 5. Steady-state exercise percentages at sea level and altitude

| | Sea Level | | Altitude, Day 3 | | Altitude, Day 10 | |
|----------------|-----------|----------|-----------------|----------|------------------|----------|
| | Low | High | Low | High | Low | High |
| Placebo | 45.6 ± 1 | 65.0 ± 2 | 52.5 ± 2 | 66.9 ± 5 | 48.7 ± 2 | 69.8 ± 4 |
| CHO | 46.0 ± 1 | 64.7 ± 1 | 49.3 ± 2 | 66.9 ± 3 | 50.2 ± 2 | 67.0 ± 2 |

Values are means ± SE. Placebo = Placebo Group; CHO = carbohydrate supplement group.

Heart Rate

Figure 1 shows heart rates during the steady-state exercise segment of the endurance test for the placebo and CHO-supplemented groups. There were no between-group

differences in resting or exercise heart rates at sea level or at altitude. Overall, heart rates during exercise were higher than during rest (A, $P<0.01$), and heart rates at the higher intensity were greater than heart rates at the lower intensity (B, $P<0.01$). Resting and exercise heart rates on day 3 at altitude were higher ($**P<0.01$) than those either at sea level or day 10 at altitude. Similar reduction in heart rate for both groups during exercise from day 3 to day 10 despite being at the same work rate and percentage of VO_{2max} is consistent with both groups experiencing a similar degree of altitude acclimatization (14).

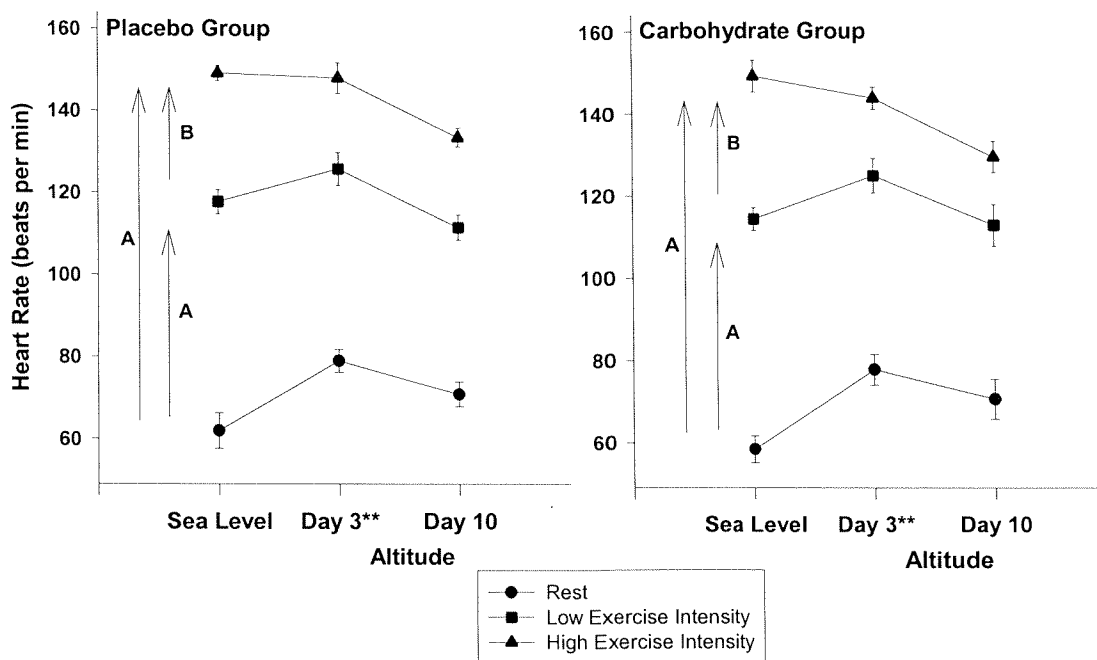


FIGURE 1: Heart Rate at Rest and During Steady-State Exercise.

% Maximal Heart Rate

Table 6 shows the percentage of maximal heart rate used during the low and high exercise intensities for each group during the steady-state exercise segment of the endurance test. For sea level and both altitude test days, percentage maximal heart rate for the low and high exercise intensities were similar for both groups. Percentages of maximal heart rates for the low and high exercise intensities were greater at altitude than for sea level for both groups ($P<0.01$), but there were no differences from day 3 to day 10 at altitude.

TABLE 6. Percentage of maximal heart rate during steady-state exercise at sea level and altitude

| | Sea Level | | Altitude, Day 3 | | Altitude, Day 10 | |
|----------------|-----------|----------|-----------------|-----------|------------------|-----------|
| | Low | High | Low | High | Low | High |
| Placebo | 62.0 ± 1 | 78.8 ± 2 | 70.9 ± 2* | 83.1 ± 2* | 70.1 ± 3* | 84.6 ± 2* |
| CHO | 61.3 ± 1 | 79.9 ± 1 | 70.9 ± 2* | 81.7 ± 1* | 72.6 ± 2* | 83.7 ± 2* |

Values are means ±SE. Placebo = Placebo Group; CHO = carbohydrate supplement group.

*P<0.01 higher than sea level

Oxygen Saturation

Figure 2 shows arterial blood saturations during the steady-state exercise segment of the endurance test for both groups. There were no between-group differences in resting or exercise oxygen saturations at sea level or at altitude. For both groups, resting and exercise blood saturations decreased from sea level to day 3 (~98% to ~78%, **P<0.01) and then increased to day 10 (~84%, P<0.01), but remained lower compared to sea level (*P<0.01). Also for both groups, on days 3 and 10 of the exposure, blood saturation during high intensity exercise was lower than during low intensity exercise (B, P<0.03). A similar increase in arterial blood saturation for both groups during rest and exercise from day 3 to day 10 despite being at the same work rate and percentage of $\text{VO}_{2\text{max}}$ on both days is consistent with both groups experiencing a similar degree of altitude acclimatization (15).

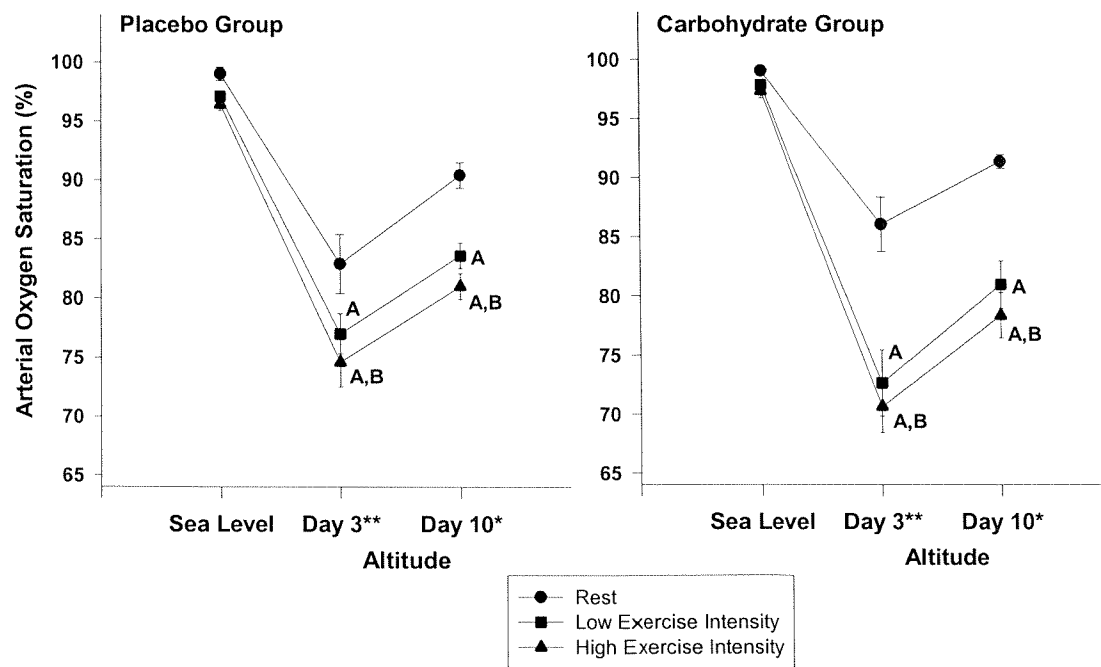


FIGURE 2: Arterial Oxygen Saturation at Rest and During Steady-State Exercise

Ratings of Perceived Exertion

There were no differences ($P > 0.05$) between groups in ratings of perceived exertion for either the low or high exercise intensities during the steady-state segment of the endurance test. For both groups, the low exercise intensity (48% $\text{VO}_{2\text{max}}$) was rated ~11 (“fairly light”) and the high exercise intensity (68% $\text{VO}_{2\text{max}}$) was rated ~14 (“somewhat hard” to “hard”) at sea level and on both days at altitude.

Blood Measures

Figures 3 to 6 show fasting blood glucose, lactate, free fatty acids, and glycerol values, respectively. Represented is the resting, and exercise values at the low (48% $\text{VO}_{2\text{max}}$) and high (68% $\text{VO}_{2\text{max}}$) exercise intensities during the steady-state exercise segment of the endurance test for both groups.

Glucose

There was no overall difference in blood glucose between groups at sea level and altitude other than a higher resting value on day 3 for the placebo group compared to CHO supplement group (102.7 ± 5 vs 96.8 ± 4 mg%, $P < 0.01$). Blood glucose was higher ($P < 0.03$)

for both groups combined for the high exercise intensity compared to resting values. Also for both groups combined, blood glucose was higher at rest (* $P < 0.01$) and during exercise (* $P < 0.01$) on day 3 at altitude compared to sea level and day 10 at altitude.

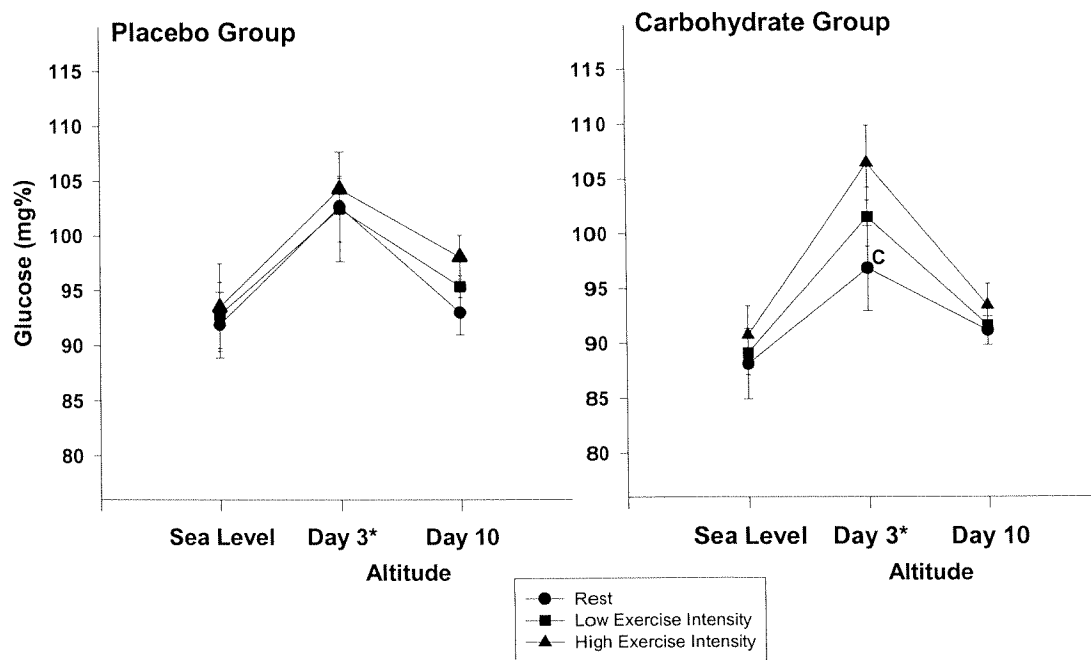


FIGURE 3: Blood Glucose at Rest and During Steady-State Exercise

Lactate

There was no overall difference in blood lactate between groups at sea level and altitude. For both groups combined, values on day 3 at altitude were higher compared to sea level and day 10 at altitude ($*P<0.01$). Lactate values were also lower on day 10 compared to sea level ($**P<0.03$). Also for both groups combined, for all test days, lactate was higher at the high exercise intensity compared to rest and the low exercise intensity ($B, P<0.01$).

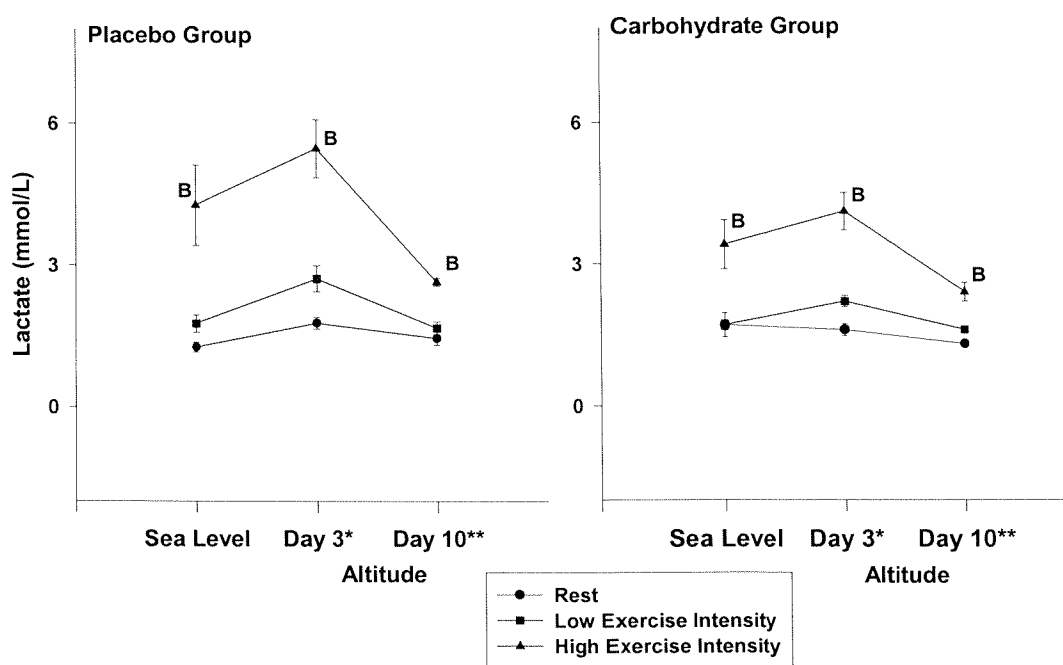


FIGURE 4: Blood Lactate at Rest and During Steady-State Exercise

Free Fatty Acids

There was no overall difference in blood free fatty acids between groups at sea level and altitude. For both groups combined, free fatty acid levels were higher on day 3 (* $P < 0.01$) and day 10 (** $P < 0.01$) at altitude compared to sea level, but did not differ between altitude days. For each test day at sea level and altitude, free fatty acid levels during exercise did not differ from rest ($P > 0.05$).

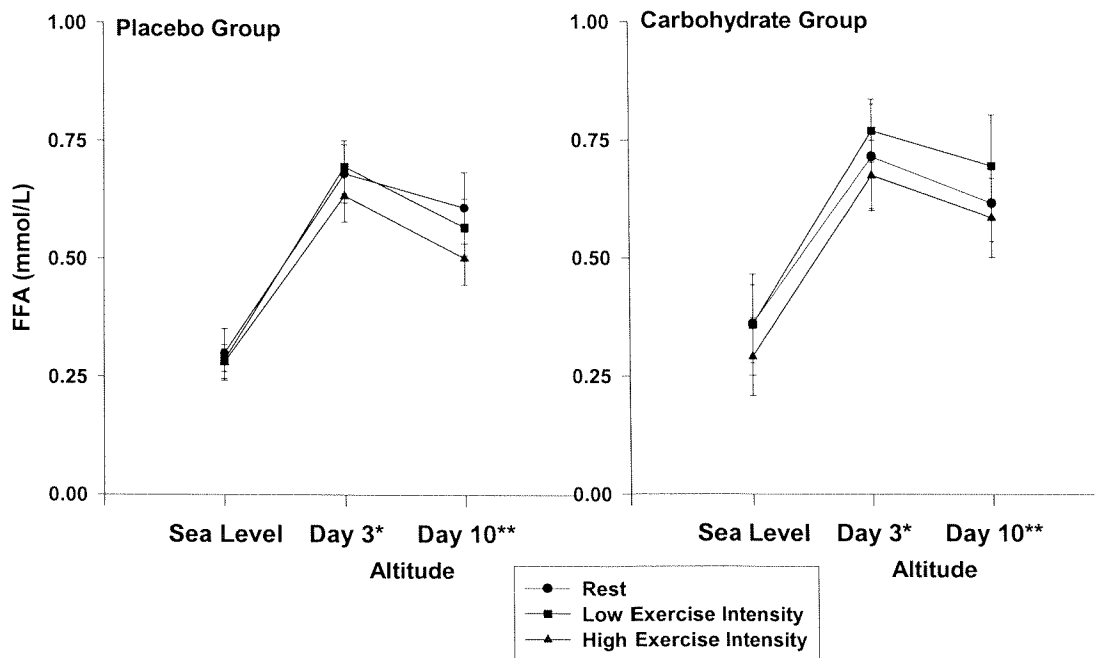


FIGURE 5: Blood Free Fatty Acid at Rest and During Steady-State Exercise

Glycerol

There was no overall difference in blood glycerol between groups at a sea level and altitude. For both groups combined, blood glycerol levels were higher at altitude than at sea level (* $P < 0.01$). Also for both groups combined and for each test day, blood glycerol was higher during both exercise intensities compared to rest (A, $P < 0.01$), and was higher during high intensity exercise compared to low intensity exercise (B, $P < 0.01$).

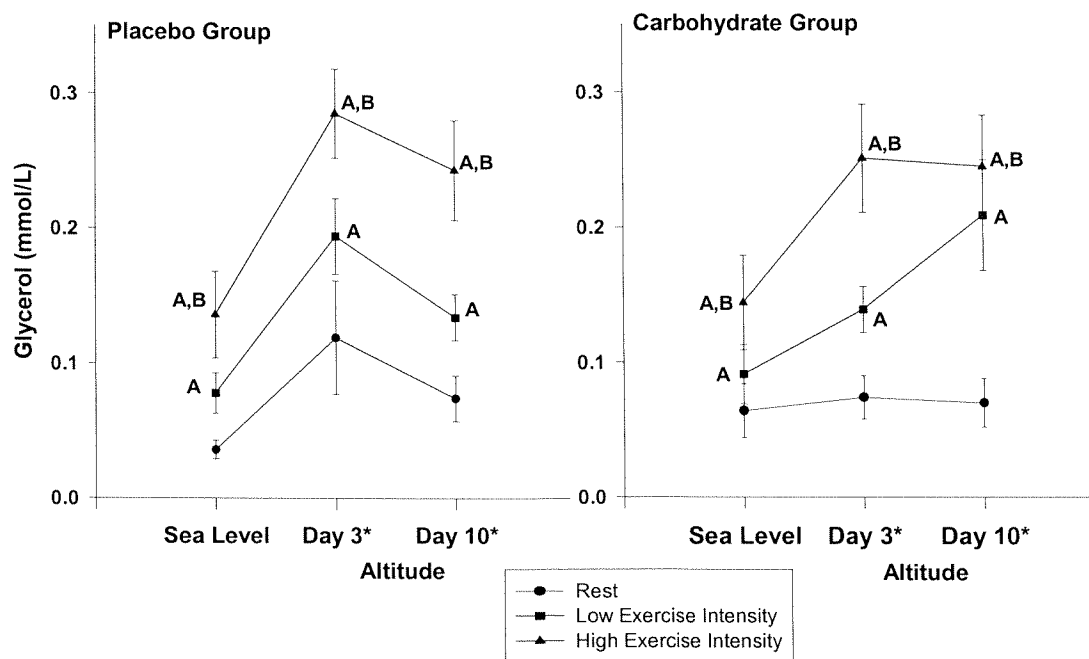


FIGURE 6: Blood Glycerol at Rest and During Steady-State Exercise

SUMMARY: STEADY-STATE EXERCISE SEGMENT

At sea level, individuals were divided into two groups that were matched on $\text{VO}_{2\text{max}}$, height, weight, and percentage body fat. In addition, during the steady-state segment of the endurance tests, both groups were fasting (i.e., no carbohydrate supplement or placebo had yet been provided). It was therefore anticipated that both groups would exercise at a similar work rate, percentage of $\text{VO}_{2\text{max}}$, and percentage of maximal heart rate. It was also anticipated that both groups during rest or exercise at sea level and altitude would have similar heart rate, arterial oxygen desaturation, and ratings of perceived exertion responses, as well as blood levels of glucose, lactate, free fatty acid, and glycerol. Except for a couple of minor exceptions, our results indicate both groups were similar and responded similarly at sea level and altitude during rest and to the steady-state segment of the endurance tests. Other results (e.g., SaO_2 and heart rate) also indicated that both groups achieved a similar degree of altitude acclimatization.

TIME-TRIAL PERFORMANCE SEGMENT

Time-Trial Performance Times

Shown in **Figure 7** are the results of the time-trial performance test during the stabilization and baseline phases at sea level and during days 3 and 10 of the altitude exposure. For both phases at sea level, there were no statistically significant differences between groups in time to complete the performance test ($P > 0.05$). Each of the groups also performed similarly between the two sea-level phases. During the stabilization phase, the time-trial was completed by the CHO-supplement group in 56.3 ± 5 min and by the placebo group in 60.7 ± 5 , and during the baseline phase, the time-trial was completed by the CHO-supplement group in 55.3 ± 5 min and by the placebo group in 62.0 ± 5 min. The coefficient of variation (CV, standard deviation / mean) from the stabilization phase to the baseline phase was 5.2% for the CHO-supplement group and 4.9% for the placebo group. These values are close to the 3.4% CV previously reported for trained cyclists completing time-trials of similar durations (18). These results clearly indicate that the test-retest variation in time-trial performance was small for both groups and that CHO supplementation during exercise had no effect on time-trial performance at sea level.

Performance times were impaired (i.e., completion time was longer) for both groups combined (A, $P<0.01$) on days 3 and 10 at altitude compared to sea level. Also for both groups combined, performance time was improved on day 10 compared to day 3 ($P<0.01$). The data also indicate that the performance time on day 3 at altitude for the placebo group was much longer than for the CHO supplement group (104.9 ± 9 vs 80.1 ± 7 min, $*P<0.01$). From day 3 to day 10 at altitude, performance time for the placebo group improved (B, $P<0.01$) whereas that for CHO supplement group did not improve ($P>0.05$). Despite the performance improvement of the placebo group from day 3 to day 10 at altitude, their time on day 10 was still longer (i.e., performance worse) than that of the CHO supplement group (89.8 ± 5 vs 76.5 ± 7 min, $*P<0.01$).

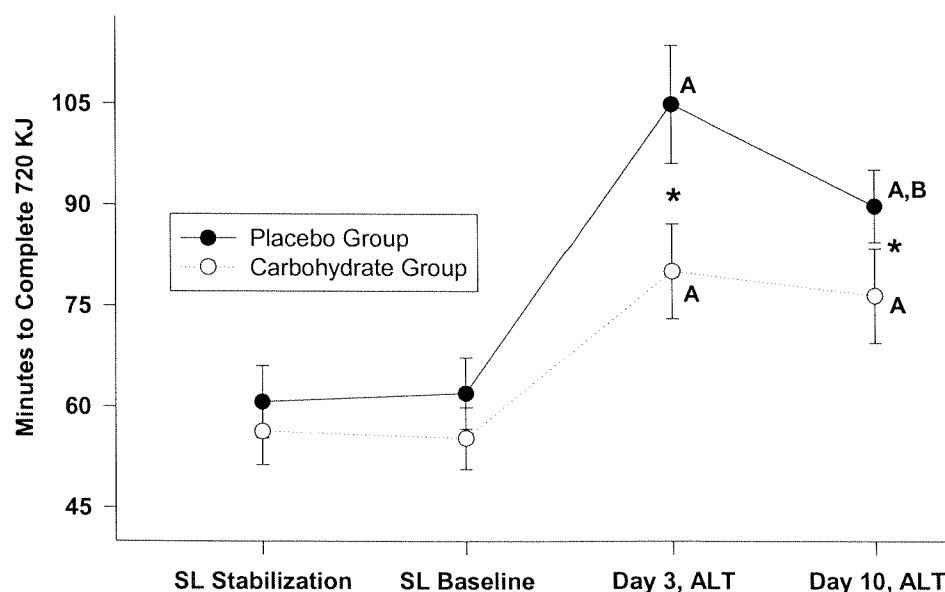


FIGURE 7: Time Trial Performance at Sea Level and Altitude

Work Rate

Shown in **Table 7** are the self-selected work rates (i.e., watts) used during the stabilization and baseline phases at sea level, and on days 3 and 10 at altitude. At sea level, each group exercised at similar work rates between the stabilization and baseline phases. Comparison of the eight pairs of volunteers matched on $\text{VO}_{2\text{max}}$ shows that those in the CHO supplement group exercised at sea level using work rates that tended to be higher (13% and 17%, $P<0.08$) than those in the placebo group. While both groups reduced absolute work rates from sea level to day 3 at altitude ($P<0.01$), the percentage difference between groups in work rate increased from ~15% at sea level to 50% on day 3 at altitude ($P<0.04$) indicating that the decline in work rate was larger for the placebo group than for the CHO group. The absolute work rates for both groups on day 10 remained lower than at sea level ($P<0.01$), but tended to be higher compared to day 3 ($P<0.07$). In addition, the percentage difference between groups in work rate was reduced at altitude from day 3 to day 10 (50% vs 25% $P=0.05$) signifying that the placebo group improved more than the CHO group. Overall, these findings are consistent with the time-trial performance times.

TABLE 7: Work Rates (Watts) at Sea Level and Altitude

| Group: | SL Stabilization | SL Baseline | Day 3, Altitude | Day 10, Altitude |
|--------------------------------|-------------------------|--------------------|------------------------|-------------------------|
| Placebo | 207 ± 16 | 203 ± 15 | 114 ± 12* | 137 ± 8* |
| Carbohydrate | 228 ± 24 | 232 ± 26 | 159 ± 15* | 167 ± 17* |
| Matched Pair Difference | 13 ± 12% | 17 ± 3% | 50 ± 20%* | 25 ± 15% ^a |

Values are means ±SE; * $P<0.01$ from sea level; ^a $P=0.05$ from day 3, altitude

% $\text{VO}_{2\text{max}}$

Shown in **Table 8** are the self-selected percentages of altitude-specific $\text{VO}_{2\text{max}}$ used during the stabilization and baseline phases at sea level, and on days 3 and 10 at altitude. Values were calculated by matching work rates during the time trial with work rates and submaximal VO_2 during the $\text{VO}_{2\text{max}}$ tests. There was no statistically significant difference in % $\text{VO}_{2\text{max}}$ used during the time-trial performance tests between sea level and day 3 at altitude

for either group. By day 10, both groups exercised at a higher %VO_{2max} compared to sea level (P<0.03) and day 3 (P<0.01).

The difference in %VO_{2max} used during the time-trial tests for the eight matched pairs of CHO supplement and placebo volunteers was ~3% at sea level but increased to 14% on day 3 at altitude (P<0.03). By day 10, the difference between groups in %VO_{2max} tended to be lower than day 3 (P=0.06) and similar to sea level. Close inspection of the data shows that the increase in percentage difference between groups at altitude was due to the CHO group using a relatively higher %VO_{2max} during the time-trial performance tests compared to the placebo group. Overall, these findings are consistent with the time-trial performance times.

TABLE 8: Percentage VO_{2max} During The Time-Trial Performance Segment

| Group: | SL Stabilization | SL Baseline | Day 3, Altitude | Day 10, Altitude |
|-------------------------|------------------|-------------|----------------------|-------------------------|
| Placebo | 61.3 ± 4 | 60.3 ± 4 | 55.1 ± 5 | 66.0 ± 3 ^{*,a} |
| Carbohydrate | 65.2 ± 3 | 64.9 ± 3 | 66.9 ± 3 | 71.8 ± 4 ^{*,a} |
| Matched Pair Difference | 4 ± 4% | 2 ± 5% | 14 ± 5% [*] | 6 ± 6% ^b |

Values are means ±SE. ^{*}P<0.03 from sea level; ^aP<0.01 from day 3; ^bP=0.06 from day 3

Heart Rate

Figure 8 shows heart rate during exercise after 25%, 50%, 75%, and 100% completion of the time-trial performance segment for both groups at sea level, and days 3 and 10 of altitude exposure. For both groups, heart rate fell progressively from sea level to days 3 (^{*}P<0.01) and 10 (^{**}P<0.01) at altitude and increased in proportion to exercise duration (A, P<0.01). For the entire study, the CHO supplement group during the time-trial performance segment tended to have higher exercise heart rates than the placebo group (P<0.08).

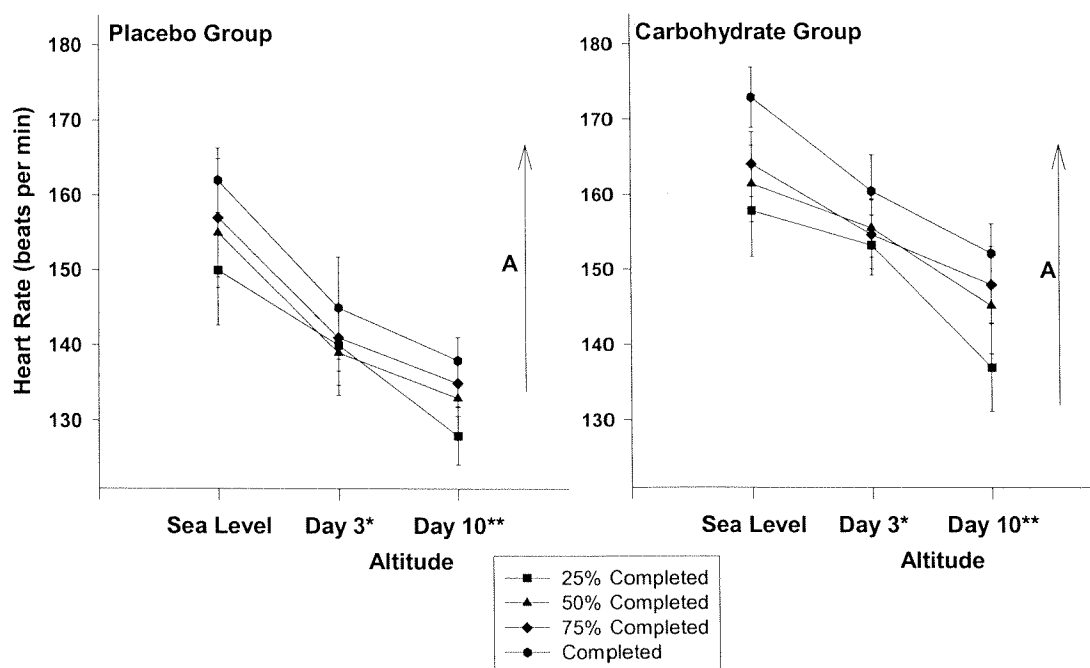


FIGURE 8: Exercise Heart Rate During the Time Trial

% Maximal Heart Rate

Figure 9 shows the percentage of altitude-specific maximal heart rate used during exercise after 25%, 50%, 75%, and 100% completion of the time-trial segment for both groups and sea level, and days 3 and 10 of altitude exposure. Values were calculated by matching time-trial heart rates with submaximal heart rates during the $\text{VO}_{2\text{max}}$ tests. Overall, the CHO supplement group used a higher percentage of maximal heart rate than the placebo group ($^{++}P<0.01$). For both groups, a higher percentage of maximal heart rate was used on day 10 of altitude compared to sea level and day 3 of altitude ($^{**}P<0.01$). Also for both groups, for all test days (with the exception of the placebo group on day 3), the percentage of maximal heart rate used was higher towards the end of exercise compared to the beginning of exercise ($P<0.01$).

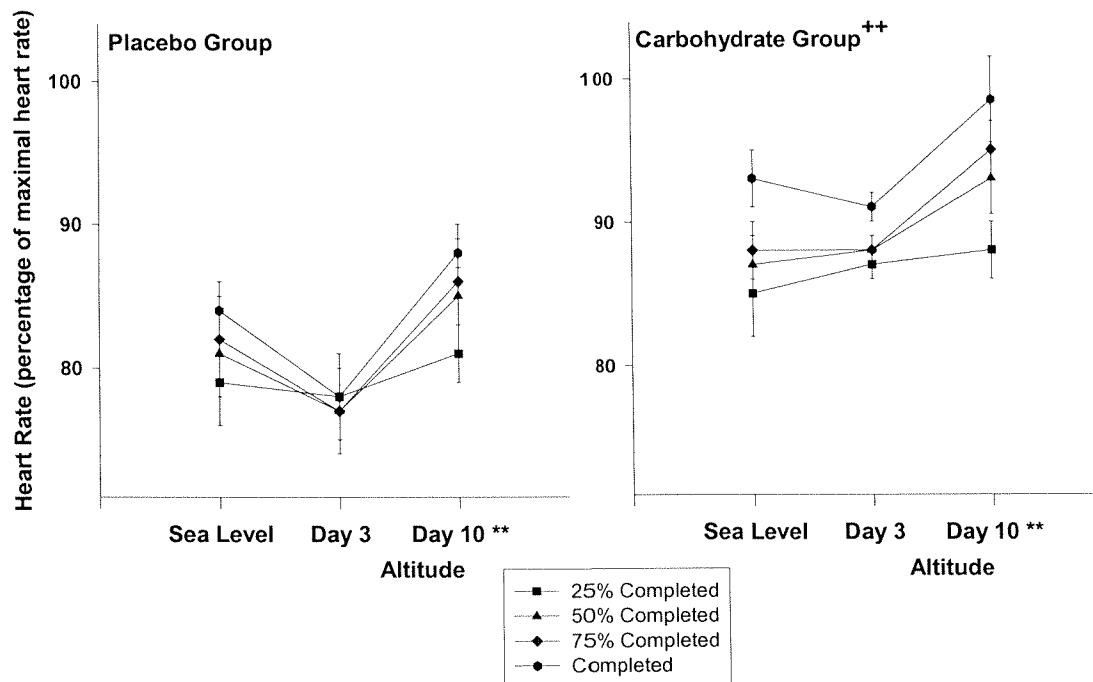


FIGURE 9: Percentage of Maximal Heart Rate Used During the Time Trial

Oxygen Saturation

Figure 10 shows arterial oxygen saturation (SaO_2) at rest, and after 25%, 50%, 75%, and 100% completion of the time-trial performance segment for both groups at sea level, and days 3 and 10 of altitude exposure. For both groups, SaO_2 during rest and exercise was lower at altitude on days 3 and 10 compared to sea level ($*P<0.01$), and was higher on day 10 compared to day 3 ($**P<0.01$). Overall, SaO_2 at altitude was lower for the CHO supplement group than for the placebo group (A, $P<0.01$) after 50% completion of the time trial performance tests. These results clearly are not consistent with the postulate that a greater degree of exercise-induced hypoxemia during CHO supplementation limits exercise performance.

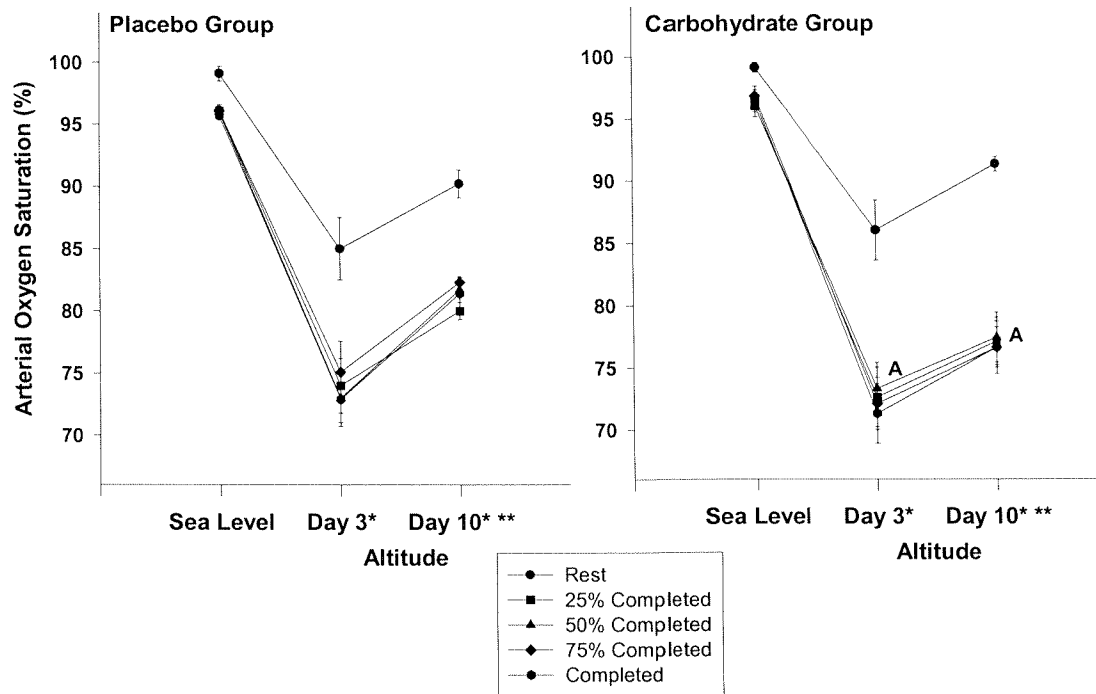


FIGURE 10: Oxygen Saturation During the Time Trial

Ratings of Perceived Exertion

Figure 11 shows ratings of perceived exertion (RPE) during exercise after 25%, 50%, 75%, and 100% completion of the time-trial performance segment for both groups at sea level, and days 3 and 10 of altitude exposure. For both groups at sea level and altitude, RPE increased progressively with exercise duration (A, $P < 0.05$). For each group, RPE did not differ among test days ($P > 0.05$). However, at all time periods after 25% completion of the time-trial, the CHO supplement group reported higher ratings of perceived exertion than the placebo group at altitude ($^{**}P < 0.01$).

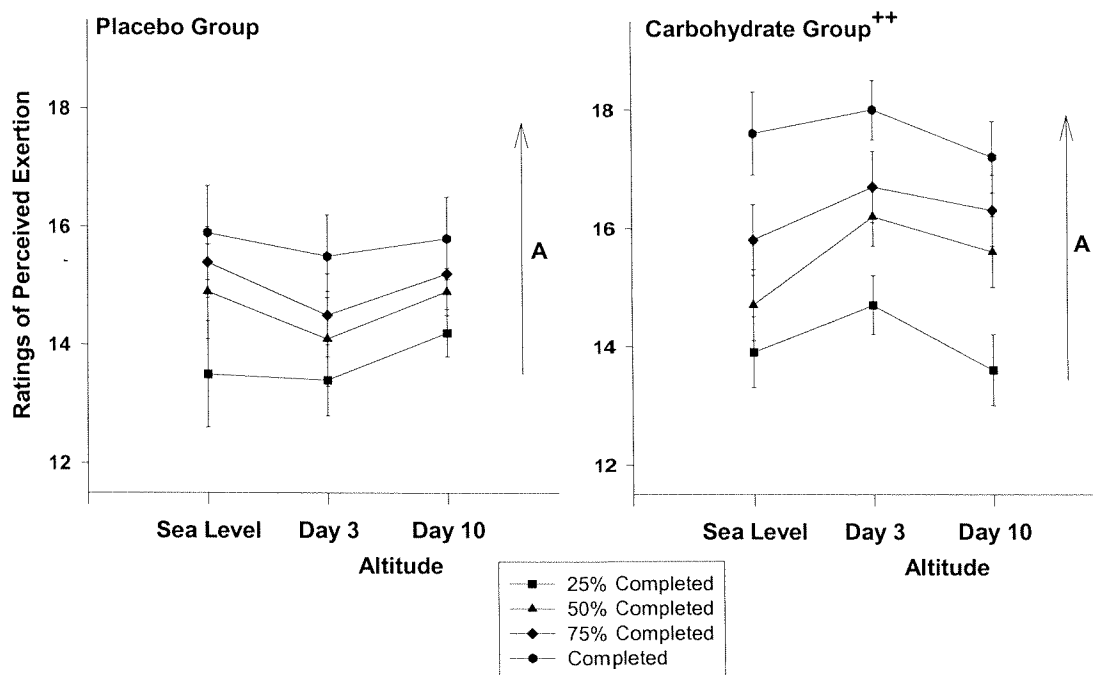


FIGURE 11: Ratings of Perceived Exertion During the Time Trial

Blood Measures

Figures 12 to 15 show blood glucose, lactate, free fatty acids, and glycerol values, respectively, for the placebo and CHO-supplemented groups during the time-trial performance segment of the endurance test. Represented are the fasting, pre-exercise resting values and exercise values after 25%, 50%, and 100% completion of the time-trial performance tests at sea level and altitude.

Glucose

At sea level and altitude, blood glucose levels during exercise were higher for the CHO group than for the placebo group ($^{++}P<0.03$). For the three test sessions, blood glucose levels for the placebo group did not differ from rest ($P>0.05$) whereas for CHO supplement group, blood glucose levels were always higher throughout exercise compared to rest (A, $P<0.02$). For both groups combined, blood glucose levels were higher on day 3 at altitude compared to sea level and day 10 at altitude ($^{*}P<0.01$). In addition, blood glucose levels were higher on day 10 at altitude compared to sea level ($^{**}P<0.01$).

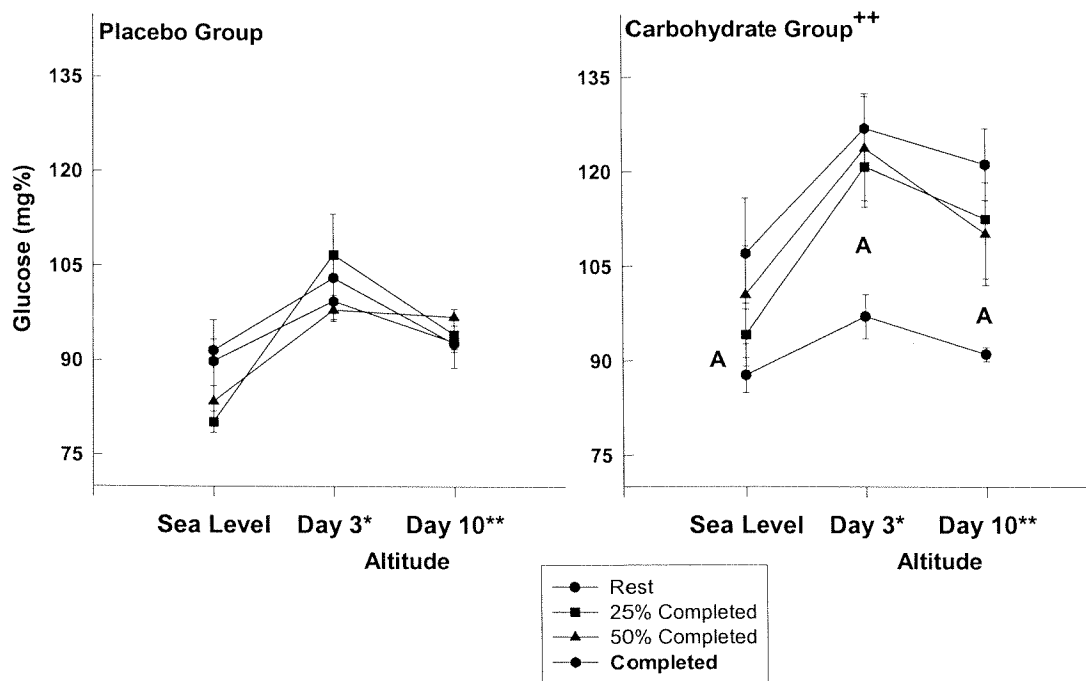


FIGURE 12: Blood Glucose During the Time Trial

Lactate

At sea level and altitude, blood lactate levels were higher for the CHO supplement group than for the placebo group ($^{++}P<0.04$). For both groups combined, blood lactate levels during exercise were similar at sea level and day 3 at altitude, but were lower on day 10 at altitude ($^{**}P<0.01$). While blood lactate levels tended to rise with increasing exercise duration (A, $P<0.07$), those for the CHO supplement group rose to higher levels compared to those of the placebo group ($P<0.03$).

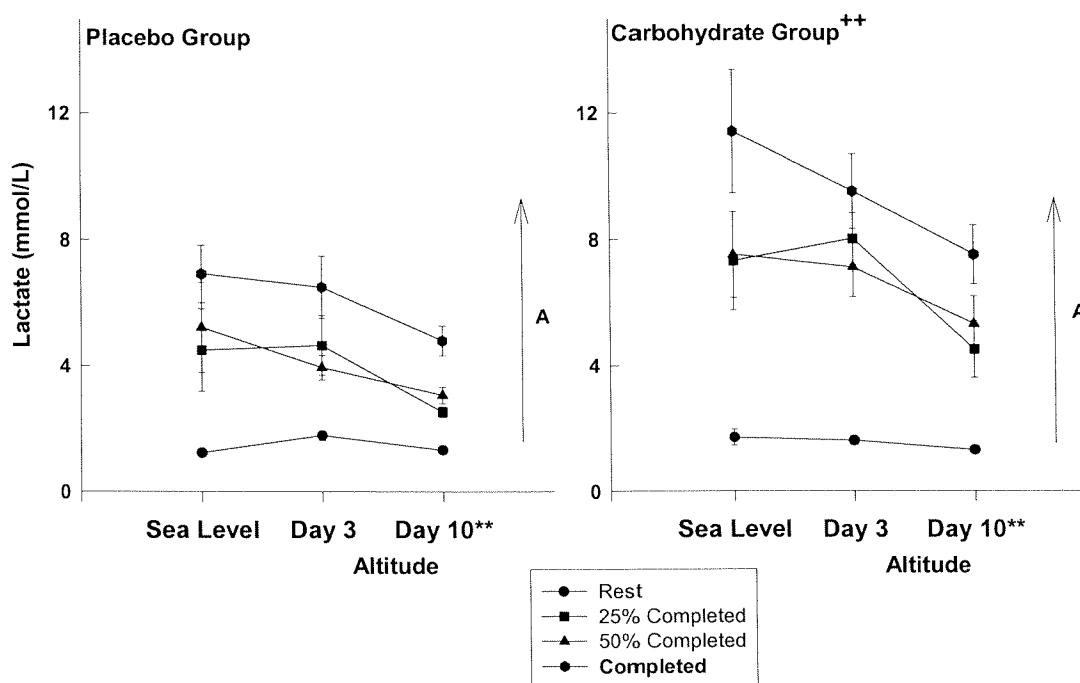


FIGURE 13: Blood Lactate During the Time Trial

Free Fatty Acids

At sea level and altitude, blood FFA levels for the placebo group were higher than for the CHO supplement group ($^{++}P<0.01$). For both groups combined, blood FFA levels were higher on day 3 at altitude compared to sea level and day 10 at altitude ($^{*}P<0.01$). Also for both groups combined, blood FFA levels were highest at the end of exercise compared to

rest and 25% and 50% completion ($P<0.01$). However, at sea level and altitude, FFA levels for the placebo group were higher during exercise compared to rest (A, $P<0.01$) in direct contrast to the CHO supplement group whose levels were lower during exercise compared to rest (B, $P<0.01$). The difference between groups in response to exercise resulted in the placebo group having higher levels of FFA during exercise than the CHO group ($P<0.01$).

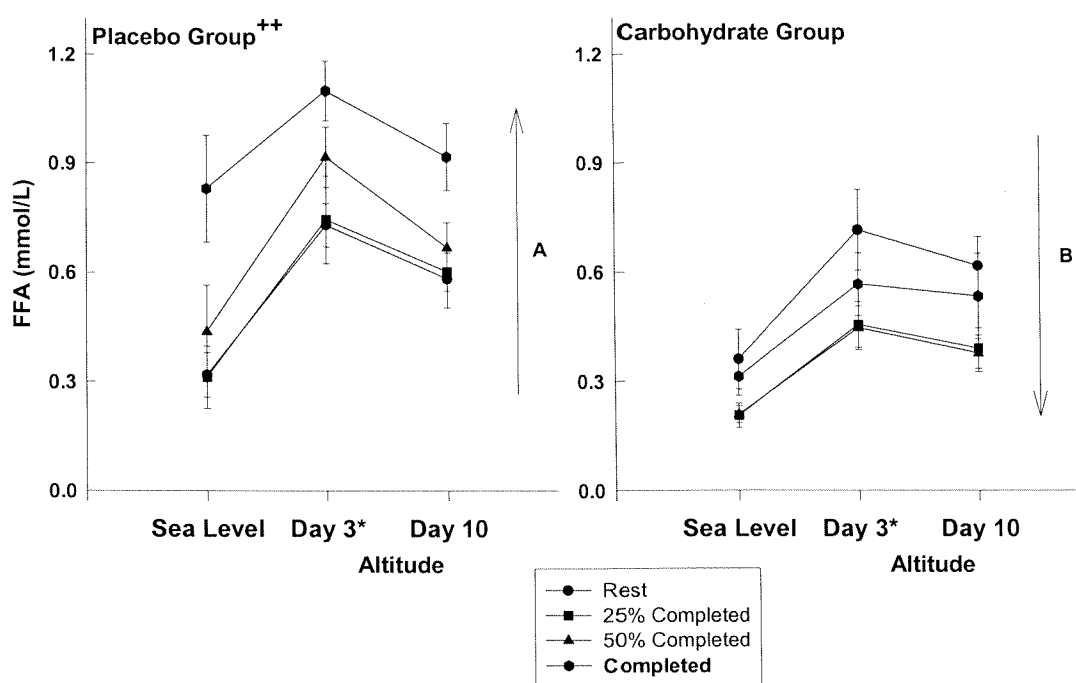


FIGURE 14: Blood Free Fatty Acids During the Time Trial

Glycerol

There was no overall difference in blood glycerol levels between groups during rest or exercise at sea level or altitude. For both groups combined, blood glycerol levels were higher at altitude than at sea level ($*P<0.01$), and, for each test day, higher during exercise than at rest (A, $P<0.01$). While blood glycerol levels rose with increasing exercise duration (B,

$P < 0.04$), those for the placebo group rose to higher levels compared to those of the CHO supplement group ($P < 0.01$).

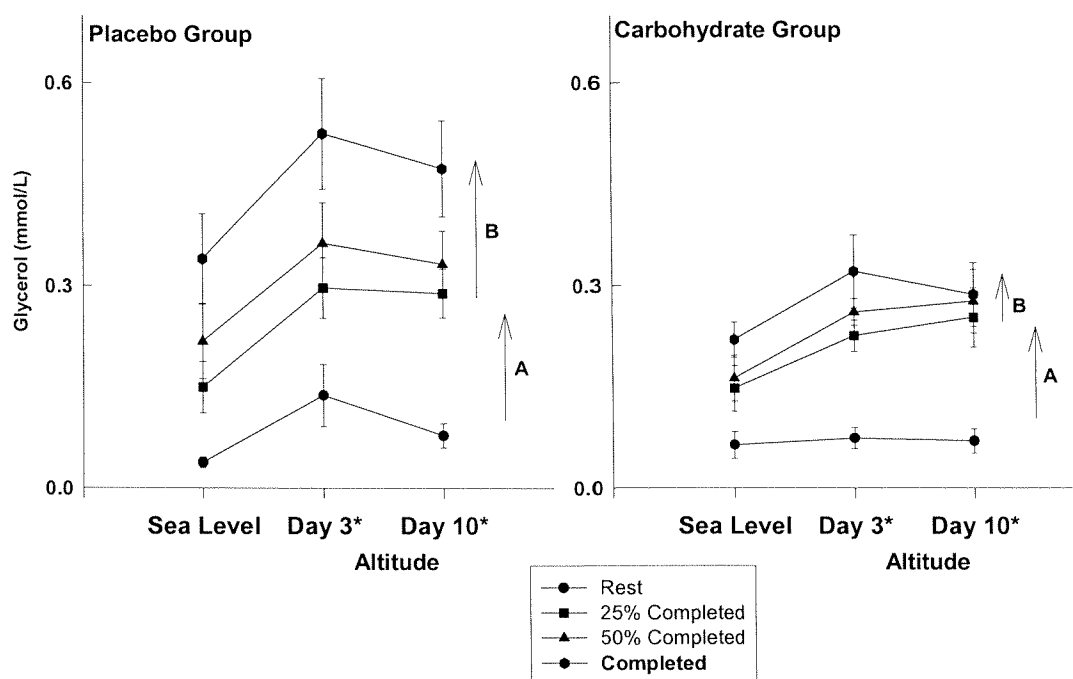


FIGURE 15: Blood Glycerol During the Time-Trial

DISCUSSION

This study is the first to show that time-trial cycle performance is greatly enhanced by CHO supplementation during exercise compared to placebo on days 3 and 10 of residence at 4,300 m. Our results are consistent with previous findings indicating that higher exercise intensities can be maintained by CHO supplementation at sea level (21; 23) and that a shift towards increased utilization of oxygen efficient fuels (i.e., glucose) occurs under conditions of acute and chronic altitude exposures (26). Moreover, while both groups performed worse at altitude than at sea level, the dominating performance of the CHO group relative to that of the placebo group on both test days at altitude despite only the placebo group improving from day 3 to day 10 is consistent with the postulate that the time-trial performance of the CHO group may have been optimized at altitude.

Carbohydrate supplementation during prolonged intense cycle exercise at sea level enhances glucose availability and oxidation, and performance at higher work rates compared to control (8; 11). However, it was not known if increased availability and oxidation of glucose could offset potential performance impairments linked to hypoxemia exacerbated by exercise at altitude. Exercise-induced hypoxemia is due to a pulmonary diffusion limitation secondary to a shorter pulmonary capillary mean transit time that results from an increase in cardiac output during exercise at altitude (28; 31). Previous reports at altitude have indicated that the higher the exercise intensity, the greater the hypoxemia (30). We therefore hypothesized that during the use of a self-paced performance paradigm at altitude, any voluntary attempt to increase exercise intensity (via CHO supplementation) would cause greater hypoxemia and hyperventilation. Such responses might be expected to limit exercise time at the higher intensity. The result would be little or no improvement in time-trial performance compared to placebo. Our data clearly do not support this postulate. In fact, the inconsistency of the relationship of SaO_2 and time-trial performance among groups for both days at altitude suggests that the magnitude of exercised-induced hypoxemia is inconsequential relative to benefit resulting from CHO supplementation. For example, for both groups there was a similar large decline in SaO_2 from rest to exercise on day 3 at altitude (~13%); yet time-trial exercise performance for the CHO group was far superior to that of the placebo group. On day 10 at altitude, the decline in SaO_2 was greater for the CHO supplement group (~14%) than for the placebo group (~9%), but time-trial exercise performance was significantly better for the CHO group. Therefore, capability of the CHO group to perform at a higher work rate and exercise intensity at altitude than the placebo group appears to be independent of the level of sustained hypoxemia and, under conditions of the present study, therefore most likely associated with increased carbohydrate availability and enhanced oxidation, like at sea level (8; 24).

At least four major experimental design and measurement outcome features of this study provide assurance that such a difference in performance at altitude between groups was related directly to CHO supplementation during exercise. First, matching groups on sea level $\text{VO}_{2\text{max}}$ minimized the possibility that the performance disparity could be ascribed to differences in fuel utilization linked to a difference in fitness level (6). Moreover, the similar altitude-induced percentage decline in $\text{VO}_{2\text{max}}$ and fall in maximal heart rate from sea level to altitude indicated that the aerobic capacity of both groups was equally affected by altitude exposure. Secondly, both groups began the steady state segment of the endurance tests

after an overnight fast in order to minimize the acute effects of the last meal on exercise metabolism and performance. Thirdly, there were striking similarities of physiological (e.g., heart rate and arterial oxygen saturation), perception (e.g., RPE), and metabolic (e.g., blood glucose and lactate) responses to steady state exercise on each testing day at altitude. Such results strongly suggest that there was a similar rate of altitude acclimatization for both groups. And fourthly, experimental bias in favor of the CHO group was avoided by having a disinterested third party make the volunteer assignments to each group. Results of the group assignments were not revealed to the other staff or volunteers until the entire study was completed. Moreover, while at altitude, volunteers from both groups also lived together, participated jointly in many of the same activities, and ate the same foods.

Previous studies at sea level reported that when resting muscle glycogen stores are replete, the rate of total CHO oxidation is similar for the CHO supplement and placebo or control groups for about 1½ to two hours of cycling at ~70% $\text{VO}_{2\text{max}}$ (12). After this time, with no CHO supplementation, muscle glycogen stores become about 60% lower and plasma glucose levels decline relative to resting levels (7; 11). Throughout the sea-level phase of the present study, energy intake nearly equaled energy expenditure and the diet was high in CHO (i.e., 63%). This suggests that at the start of the endurance performance tests for both groups, resting muscle glycogen stores were not greatly reduced from fully replete levels. In addition, each group exercised at 60 to 65% $\text{VO}_{2\text{max}}$ for less than a total of 1.5 hours (i.e., 20 min at the high exercise intensity during the steady-state segment and time-trial segment combined), and, for the placebo group, blood glucose was maintained at resting levels throughout the endurance test. Such a scenario suggests that CHO availability would not be limiting to performance, and CHO supplementation during exercise would not be expected to greatly enhance exercise performance (3; 13). The lack of change in the intra- and inter-group performance at sea level in the present study is consistent with this interpretation.

Maximal oxygen consumption at altitude was reduced from sea level for both groups and, as a result, the self-selected power outputs (i.e., watts) that could be used during the time-trial performance tests also had to be reduced. Since the total amount of work that was performed at sea level and altitude was identical (i.e., 720 KJ), the duration of the time trial was necessarily longer at altitude than at sea level for both groups. However, in contrast to the similarity in performance between groups at sea level, the performance of the CHO supplement group was far superior to that of the placebo group on both test days at altitude. It appears there are several factors that when combined could at least partly explain the time

trial performance difference at altitude. One factor for the difference between groups at altitude when none existed at sea level may be related to the observed sea level versus altitude difference in energy balance. At sea level, both groups were presumably in energy balance whereas at altitude both groups had greatly increased energy expenditures and reduced energy intakes amounting to a daily energy deficit of approximately 1450 kcals/day. Significantly increasing daily energy expenditure increased muscle glycogen utilization and, with little time for rest at altitude, likely did not allow sufficient time for muscle glycogen stores to fully recover (19). In addition, the reduced daily energy intake may not have included adequate CHO for full muscle glycogen repletion (10). It should be noted that under conditions of reduced CHO stores prior to the onset of exercise, CHO supplementation during exercise could improve exercise performance in events even shorter than about an hour (24).

In addition to the likelihood that both groups started the time-trial performance test at altitude with less than fully replete resting muscle glycogen stores, each group exercised at approximately similar % $\text{VO}_{2\text{max}}$ at altitude and sea level. Since muscle glycogen utilization is related more to % $\text{VO}_{2\text{max}}$ than absolute work rate (29) and the volunteers exercised for longer periods of time at altitude than at sea level, it is likely that muscle glycogen stores fell to lower levels at altitude than at sea level. The implication is that as muscle glycogen progressively declines during prolonged exercise, the reliance on blood glucose to maintain total CHO oxidation steadily increases (8). In the present study, there was no decline in blood glucose in the placebo group during exercise compared to rest. Blood glucose levels for the CHO group were, however, higher than for the placebo group at sea level and altitude, and performance was superior for the CHO group at altitude. These results are consistent with previous findings indicating that during exercise at altitude skeletal muscle becomes more dependent on glucose, an oxygen-efficient fuel (5; 25; 26) and is consistent with the postulate that under such conditions increasing glucose availability increased glucose utilization and resulted in improved exercise performance.

SUMMARY

Two groups of eight men, matched by age, $\text{VO}_{2\text{max}}$, body weight, and height, performed cycle endurance tests at sea level and on days 3 and 10 while living at 4300 m altitude. The endurance test consisted of two distinct segments: a steady-state exercise

segment (i.e., 20 min at 48% and 68% $\text{VO}_{2\text{max}}$) that assessed physiological changes due to altitude acclimatization while fasting and a 720 KJ time trial segment that assessed the effects of CHO supplementation (10% solution, 0.7 g/kg bw, every 15 min) on prolonged, maximal effort performance. During the endurance tests, heart rate, arterial oxygen saturation, ratings of perceived exertion, and blood levels of glucose, lactate, free fatty acids, and glycerol were measured repeatedly. Both groups had similar physiological responses to the steady-state exercise segment at sea level and altitude, and acclimatized similarly with altitude residence. In contrast, time-trial exercise performance at altitude was far superior for the CHO group than for the placebo group despite severe exercise-hypoxemia. We conclude that increased availability of glucose as a metabolic fuel can greatly improve endurance performance at altitude.

REFERENCES

1. **Ainsworth BE, Haskell WL, Leon AS, Jacobs JrDR, Montoye HJ, Sallis JF and Paffenbarger JrRS.** Compendium of physical activities: Classification of energy costs of human physical activities. *Med Sci Sports Exerc* 25: 71-80, 1993.
2. **American College of Sports Medicine, American Dietetic Association and Dietitians of Canada.** Nutrition and athletic performance. *Med Sci Sports Exerc* 2130-2145, 2000.
3. **Bjorkman O, Sahlin K, Hagenfeldt L and Wahren J.** Influence of glucose and fructose ingestion on the capacity for long-term exercise. *Clin Physiol* 4: 483-494, 1984.
4. **Borg GAV.** Psychophysical basis of perceived exertion. *Med Sci Sports Exerc* 14: 377-381, 1982.
5. **Brooks GA, Butterfield GE, Wolfe RR, Groves BM, Mazzeo RS, Sutton JR, Wolfel EE and Reeves JT.** Increased dependence on blood glucose after acclimatization to 4,300 m. *J Appl Physiol* 70: 919-927, 1991.
6. **Brooks GA and Mercier J.** The balance of carbohydrate and lipid utilization during exercise: the "crossover" concept. *J Appl Physiol* 76: 2253-2261, 1994.
7. **Coggan AR and Coyle EF.** Reversal of fatigue during prolonged exercise by carbohydrate infusion or ingestion. *J Appl Physiol* 63: 2388-2395, 1987.
8. **Coggan AR and Coyle EF.** Carbohydrate ingestion during prolonged exercise: Effects on metabolism and performance. In: *Exercise and Sport Sciences Reviews*, edited by Hollosky JO. Philadelphia: Pa, 1991, p. 1-40.

9. **Costill DL.** Carbohydrates for exercise: dietary demands for optimal performance. *Int J Sports Med* 9: 1-18, 1988.
10. **Costill DL, Flynn MG, Kirwan JP, Houmard JA, Mitchell JB, Thomas R and Han Park S.** Effects of repeated days of intensified training on muscle glycogen and swimming performance. *Med Sci Sports Exerc* 20: 249-254, 1988.
11. **Coyle EF.** Timing and method of increased carbohydrate intake to cope with heavy training, competition and recovery. In: *Foods, Nutrition and Sports Performance*, edited by Williams C and Devlin JT. London: E and FN SPON, 1992, p. 35-63.
12. **Coyle EF, Coggan AR, Hemmert MK and Ivy AC.** Muscle glycogen utilization during prolonged strenuous exercise when fed carbohydrate. *J Appl Physiol* 61: 165-172, 1986.
13. **Coyle EF, Hagberg JM, Hurley BF, Martin WH, Ehsani AA and Hollosky JO.** Carbohydrate feedings during prolonged strenuous exercise can delay fatigue. *J Appl Physiol* 55: 230-235, 1983.
14. **Fulco CS, Rock PB and Cymerman A.** Maximal and submaximal exercise performance at altitude. *Aviat Space Environ Med* 69: 793-801, 1998.
15. **Grover RF, Weil JV and Reeves JT.** Cardiovascular adaptation to exercise at high altitude. In: *Exercise and Sport Science Reviews*, edited by Pandolf KB. New York: Macmillan, 1986, p. 269-302.
16. **Harris JA and Benedict FG.** *A Biometric Study of Basal Metabolism in Man.* Washington, D.C.: Carnegie Institution, 1919.
17. **Ivy JL, Costill DL, Fink WJ and Lower RW.** Influence of caffeine and carbohydrate feedings on endurance performance. *Med Sci in Sports* 11: 6-11, 1979.

18. **Jeukendrup A, Saris WHM, Brouns F and Kester ADM.** A new validated endurance performance test. *Med Sci Sports Exerc* 28: 266-270, 1996.
19. **Kirwan JP, Costill DL, Mitchell JB, Houmard JA, Flynn MG, Fink WJ and Belur ER.** Carbohydrate balance in competitive runners during successive days of intense training. *J Appl Physiol* 65: 2601-2606, 1988.
20. **McGrane MM.** Carbohydrate Metabolism. Synthesis and Oxidation. In: Biochemical and Physiological Aspects of Human Nutrition, edited by Stipanuk MH. Philadelphia: W.B. Saunders Company, 2000, p. 158-210.
21. **Mitchell JB, Costill DL, Houmard JA, Fink WJ, Pascoe DD and Pearson DR.** Influence of carbohydrate dosage on exercise performance and glycogen metabolism. *J Appl Physiol* 67: 1843-1849, 1989.
22. **Montain, S. J., McGraw, S. M., Tharion, W. J., Kramer, F. M., and Hoyt, R. W.** Acceptability of carbohydrate drinks during the Marine Corps infantry officer course 10-day field training exercise. U.S.Army Research Institute of Environmental Medicine T-00-2, AD A-370960. 1999.
23. **Murray R, Eddy DE, Murrar TW, Seifert JG, Paul GL and Halaby GA.** The effect of fluid and carbohydrate feedings during intermittent cycling exercise. *J Appl Physiol* 19: 597-604, 1987.
24. **Neufer DP, Costill DL, Flynn MG, Kirwan JP, Mitchell JB and Houmard JA.** Improvements in exercise performance: effects of carbohydrate feedings and diet. *J Appl Physiol* 62: 983-988, 1987.

25. **Roberts AC, Butterfield GE, Cymerman A, Reeves JT, Wolfel EE and Brooks GA.** Acclimatization to 4,300-m altitude decreases reliance on fat as a substrate. *J Appl Physiol* 81: 1762-1771, 1996.
26. **Roberts AC, Reeves JT, Butterfield GE, Mazzeo RS, Sutton JR, Wolfel EE and Brooks GA.** Altitude and B-blockade augment glucose utilization during submaximal exercise. *J Appl Physiol* 80: 605-615, 1996.
27. **Rowell LB.** Cutaneous and skeletal muscle circulations. In: *Human Circulation: Regulation during Physical Stress*, edited by Rowell LB. New York: Oxford University Press, 1986, p. 96-116.
28. **Rowell LB, Saltin B, Kiens B and Juel Christensen N.** Is peak quadriceps blood flow in humans even higher during exercise with hypoxemia? *Am J Physiol* 251: H1038-H1044, 1986.
29. **Saltin B and Karlsson J.** Muscle glycogen utilization during work of different intensities. In: *Muscle Metabolism During Exercise*, edited by Pernow B and Saltin B. New York: Plenum, 1971, p. 289-299.
30. **Sutton JR, Reeves JT, Wagner PD, Groves BM, Cymerman A, Malconian M, Rock PB, Young PA, Walter SD and Houston CS.** Operation Everett II: oxygen transport during exercise at extreme simulated altitude. *J Appl Physiol* 64: 1309-1321, 1988.
31. **Wagner PD, Gale GE, Moon RE, Torre-Bueno JR, Stolp BW and Saltzman HA.** Pulmonary gas exchange in humans exercising at sea level and simulated altitude. *J Appl Physiol* 61: 260-270, 1986.